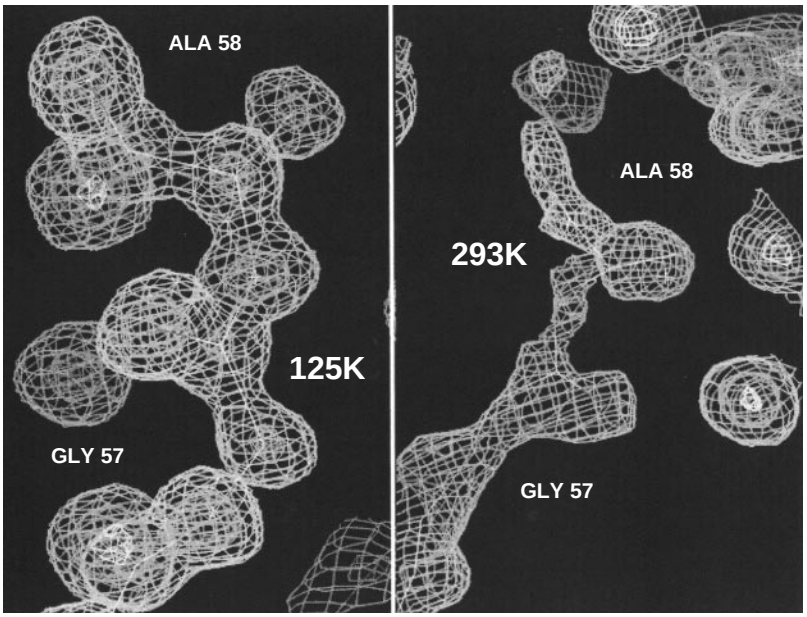
High Resolution Cryo-crystallography of Biological Macromolecules

Modern cryo-cooling techniques provide macromolecular structures of unprecedented detail

Knowledge of accurate molecular structures is a prerequisite for rational drug design and for structure based functional studies to aid the development of effective therapeutic agents. X-ray crystallography provides a powerful tool to determine the structure of large macromolecules, such as proteins, DNA oligomers, and complexes thereof.



Comparison of equivalent regions in a macromolecular structure: left, flash-cooled to 125 K; right, recorded at room temperature.

APPLICATIONS

- *Atomic resolution of protein structures*
- *Accurate molecular structure determination for rational drug design*
- *Studies of protein interactions with DNA and other proteins*
- *DNA repair*
- *Apolipoproteins*
- *Signal transduction proteins*
- *Growth factor related oncogenes*

Flash cooling improves data quality

The most promising way to achieve higher resolution and an overall increase in data quality is to flash-cool the crystals to liquid nitrogen temperature (77K). Obvious benefits are reduced thermal vibrations, enhanced signal-to-noise ratio, reduced conformational disorder, and in many cases, a higher limiting resolution. The most important effect, however, is the suppression of radiation damage, permitting a complete data set to be collected from one single crystal. This in turn eliminates errors from merging and scaling of data sub-sets from multiple crystals. In addition, crystal mounting is dramatically simplified over conventional capillary techniques.

Instrumental design improvements provide additional benefits

In addition to enhanced cryo-cooling, we have incorporated focusing mirror design, kappa-diffraction geometry, and large area detectors providing a reduction of collection time by a factor of eight. Consequently, we can obtain high quality data within a day or two compared to one to three weeks. In combination with cryo-cooling, these improvements lead to enhanced contrast and sharper detail in electron density maps, facilitating model building and reducing the total time required for structure determination. We are currently applying these cryo-techniques to apo-E lipoproteins, which play a role in lipid disorders such as Alzheimer's disease, to signal transduction proteins, and to cell growth factor related oncogenes.

Availability: The technology is available now. We welcome discussions with representatives of pharmaceutical companies to discuss how our technologies might meet their needs.

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